



**United States Environmental Protection Agency  
Region 9 Laboratory**

1337 S. 46th Street Building 201  
Richmond, CA 94804

**Date:** 5/6/2015  
**Subject:** Analytical Testing Results - Project R15N02  
SDG: 15105A  
**From:** Duane James, Acting Director  
EPA Region 9 Laboratory  
MTS-2  
**To:** Eugene E. Bromley  
NPDES Permits Section  
WTR-2-3

A handwritten signature in black ink, appearing to be "Duane James", written over the "From:" field.

Attached are the results from the analysis of samples from the **Southern California Oil Platforms Spring 2015** project. These data have been reviewed in accordance with EPA Region 9 Laboratory policy.

A full documentation package for these data, including raw data and sample custody documentation, is on file at the EPA Region 9 Laboratory. If you would like to request additional review and/or validation of the data, please contact Eugenia McNaughton at the Region 9 Quality Assurance Office.

If you have any questions, please ask for Amy Wagner, the Lab Project Manager at (510)412-2300.

Electronic CC: Dave Panzer, david.panzer@mms.gov  
Susan Zaleski, susan.zaleski@boem.gov  
James Salmons, james.salmons@bsee.gov

**Analyses included in this report:**

---

Abalone Toxicity



# United States Environmental Protection Agency Region 9 Laboratory

1337 S. 46th Street, Building 201, Richmond, CA 94804  
Phone:(510) 412-2300 Fax:(510) 412-2302

<b>Project Manager:</b> Eugene E. Bromley	<b>NPDES Permits Section</b>	<b>SDG:</b> 15105A
<b>Project Number:</b> R15N02	<b>75 Hawthorne Street</b>	<b>Reported:</b> 05/06/15 13:29
<b>Project:</b> Southern California Oil Platforms Spring 2015	<b>San Francisco CA, 94105</b>	

## ANALYTICAL REPORT FOR SAMPLES

Sample ID	Laboratory ID	Matrix	Date Collected	Date Received
Platform A	1504030-01	Water	04/14/15 08:40	04/15/15 10:50
Platform B	1504030-02	Water	04/14/15 10:50	04/15/15 10:50

**SDG ID 15105A**

### Work Order(s)

**1504030**

Requested analyses were for abalone development toxicity tests using *Haliotis rufescens* (red abalone) following Short Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to West Coast Marine and Estuarine Organisms, EPA/600/R-95/136 and USEPA Region 9 Laboratory SOP #1004, RED ABALONE (*Haliotis rufescens*) LARVAL DEVELOPMENT TOXICITY TEST, was followed for the toxicity tests. A concurrent reference toxicity test was conducted for quality control as specified in the method. Statistical analyses were conducted using the CETIS statistical database program, version 1.022, and the Test of Significant Toxicity, version 1.8.

The test concentrations were based on the oil platform NPDES general permit requirements using the Test of Significant Toxicity (TST) approach described in *National Pollutant Discharge Elimination System Test of Significant Toxicity Implementation Document* (EPA 833-R-10-003, 2010). The chronic WET permit limit that must be met is rejection of the null hypothesis (Ho). The test results rejected the null hypothesis and are reported as Pass for both Platform A and B samples.

All QA/QC criteria were met with the following exception.

1. Since the measured value of the reference toxicant was outside of 15% of the nominal value, the reference toxicant concentrations were recalculated, data were re-analyzed using statistical analyses, and data was re-plotted in the control chart.



**United States Environmental Protection Agency  
Region 9 Laboratory**

1337 S. 46th Street, Building 201, Richmond, CA 94804  
Phone:(510) 412-2300 Fax:(510) 412-2302

<b>Project Manager:</b> Eugene E. Bromley	<b>NPDES Permits Section</b>	<b>SDG:</b> 15105A
<b>Project Number:</b> R15N02	<b>75 Hawthorne Street</b>	<b>Reported:</b> 05/06/15 13:29
<b>Project:</b> Southern California Oil Platforms Spring 2015	<b>San Francisco CA, 94105</b>	

**Sample Results**

Analyte	Reanalysis / Extract	Result	Qualifiers / Comments	Quantitation Limit	Units	Batch	Prepared	Analyzed Method
<b>Lab ID:</b> 1504030-01							<b>Water - Sampled: 04/14/15 08:40</b>	
<b>Sample ID:</b> Platform A							<b>Aquatic Toxicity Test by EPA Methods</b>	
Test of Significant Toxicity		Pass	A-01		%	B15E010	04/15/15	04/15/15 TOX/SOP1004
<b>Lab ID:</b> 1504030-02							<b>Water - Sampled: 04/14/15 10:50</b>	
<b>Sample ID:</b> Platform B							<b>Aquatic Toxicity Test by EPA Methods</b>	
Test of Significant Toxicity		Pass	A-01		%	B15E010	04/15/15	04/15/15 TOX/SOP1004

**Quality Control**

Analyte	Result	Qualifiers / Comments	Quantitation Limit	Units	Spike Level	Source Result	%REC	%REC Limits	RPD	RPD Limit
<b>Batch B15E010 - - General Biology - Toxicity, Abalone</b>							<b>Prepared &amp; Analyzed: 04/15/15</b>			
<b>Reference (B15E010-SRM1)</b>							<b>Aquatic Toxicity Test by EPA Methods - Quality Control</b>			
EC 50	51.6			ppb	52.1		99	54.8-145.2		



United States Environmental Protection Agency  
**Region 9 Laboratory**

1337 S. 46th Street, Building 201, Richmond, CA 94804  
Phone:(510) 412-2300 Fax:(510) 412-2302

<b>Project Manager:</b> Eugene E. Bromley	<b>NPDES Permits Section</b>	<b>SDG:</b> 15105A
<b>Project Number:</b> R15N02	<b>75 Hawthorne Street</b>	<b>Reported:</b> 05/06/15 13:29
<b>Project:</b> Southern California Oil Platforms Spring 2015	<b>San Francisco CA, 94105</b>	

**Qualifiers and Comments**

Pass Pass

A-01 Refer to the TST worksheet for statistical analysis

U Not Detected

NR Not Reported

RE1, RE2, etc: Result is from a sample re-analysis.

**QUALITY CONTROL SUMMARY FORM**

**Abalone Development Toxicity Test**

Test numbers: 1504030-01, -02 Site: Southern CA Oil Platforms Test Date: 4/15/2015

	<u>YES</u>	<u>NO</u>	<u>COMMENTS</u>
<b>Controls</b>			
≥ 80% mean larval normality in controls	<u>X</u>	_____	_____

**Reference Toxicant Test**

Control chart attached	<u>X</u>	_____	_____
NOEC value	<u>21.06 ug/L</u>		_____

NOEC (No Observable Effect Concentration) - The highest concentration of a toxicant at which no adverse effects are observed on the test organisms (i.e., the highest concentration that is not statistically significantly different from the controls).

EC<sub>50</sub> value                      51.64                      Low Confidence Interval 50.10

High Confidence Interval 53.21

EC<sub>50</sub> (50% Effect Concentration) - The EC<sub>50</sub> is the concentration of a toxicant that would cause a 50% reduction in normal larval shell development for the test population.

% MSD < 20% relative to control	<u>X</u>	_____	<u>4.20%</u>
%MSD - A measure of the within-test variability representing the amount of difference from the control that can be detected statistically. (%MSD = MSD/control mean x 100)			

Statistically significant effect at 56 ug/L	<u>X</u>	_____	_____
---	----------	-------	-------

**Water Quality Measurements (within the acceptable ranges listed below)**

Acceptable Range

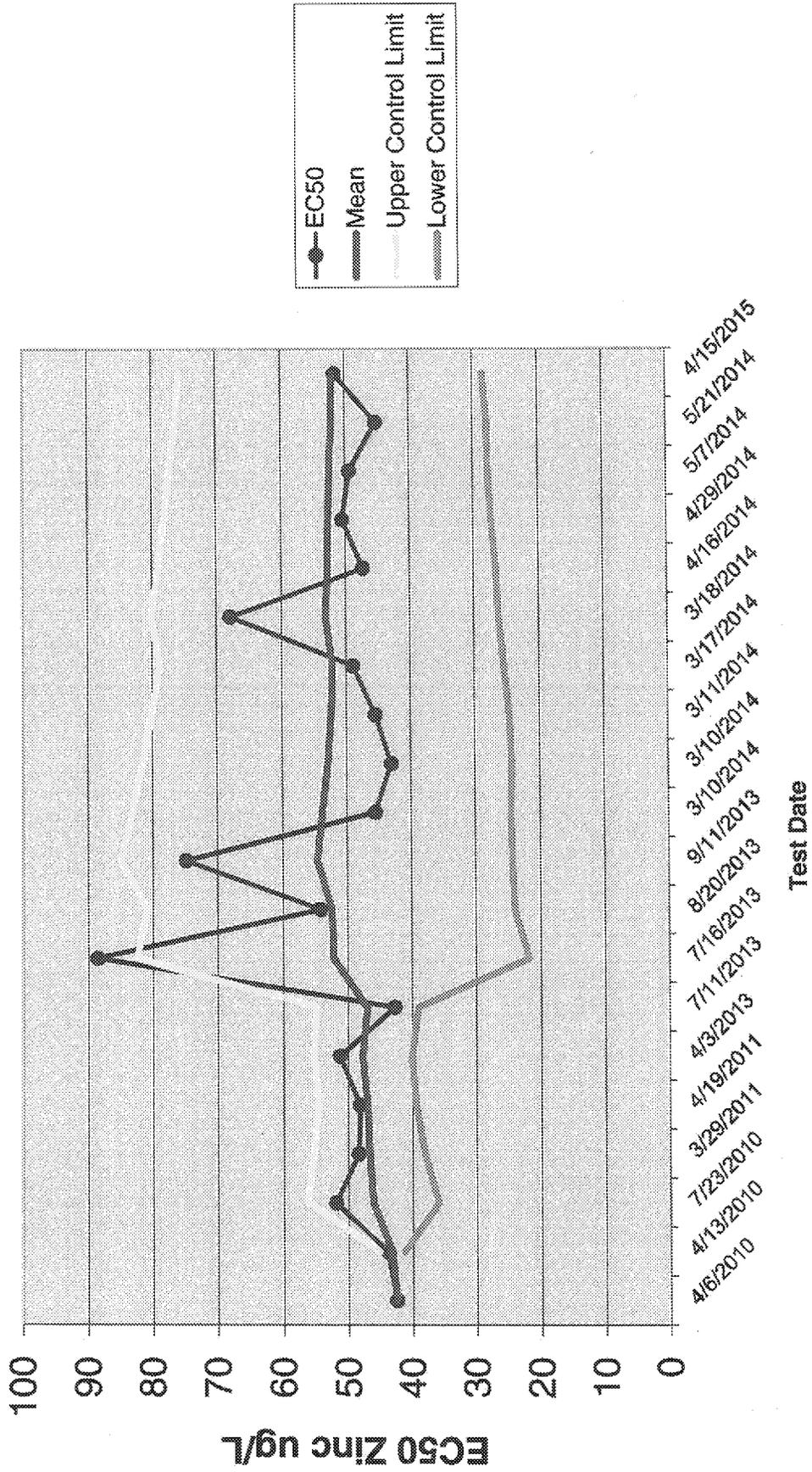
Dissolved oxygen (mg/L)	>65% sat.	<u>X</u>	_____	_____
pH (units)	7.5-8.5	<u>X</u>	_____	_____
Salinity (ppt)	32-36	<u>X</u>	_____	_____
Temperature (°C)	14-16°C	<u>X</u>	_____	_____

Name: Amy Wagner

Title: Marine Biologist



# Red Abalone Control Chart Zinc Sulfate





# TST Calculator v1.8

## TST Summary

The Test of Significant Toxicity (TST) is a new statistical approach that assesses the whole effluent toxicity (WET) measurement of wastewater effects on specific test organisms' ability to survive, grow, and reproduce developed by the U.S. EPA. TST uses hypothesis testing techniques which examines whether a sample, at the critical concentration (e.g., in-stream waste concentration or IWC), and the control within a WET test differ by an unacceptable amount (the amount that would have a measured adverse effect on the ability of aquatic organisms to thrive and survive). A sample can be either an effluent, stormwater, or receiving water. Once the WET test has been conducted, the TST approach can be used to analyze valid WET test results to assess whether the sample is toxic. The TST approach is designed to be used for a two concentration data analysis of the sample's critical concentration (e.g., IWC or a receiving water concentration) as compared to a control concentration.

## Instructions

Before using this program, Macros must be enabled and the security certificate must be accepted.  
For Excel 2007

1. Click the "Office Button" (big Office logo in the upper left), and select "Excel Options" (lower right)
2. Click "Trust Center" on the left menu
3. Click "Trust Center Settings" (lower right)
4. Click "Macro Settings"
5. Click "Enable all macros" (last option)
6. Click "ok" buttons to clear all open windows
7. Exit program and re-open the file to use

For Excel pre-2007

1. Go to "Tools", "Macro", and Click "Security"
2. Set Macro security to medium
3. Exit program and re-open file
4. Click "Enable Macros"

To use the program:

1. Enter test information in the first block of red shaded cells (Test ID, Test Date, Test Duration, and Critical Concentration)
  - \* "Critical Conc." means a facility specific IWC or 100% receiving water, etc. (i.e., one concentration of WET test)
2. Fill in your raw test data (Number of Organisms Exposed or Counted and Response) for as many replicates as conducted in WET test
  - \* "Critical Conc. Data" means data from the concentration entered as the "Critical Conc." in cell B7
  - \* Percent data (survival, germination, etc.) should be input as whole numbers (not as decimals)
  - \* Fish growth must be input as biomass
3. Choose the appropriate test information from the red "Click to choose from menu" cells
4. All gray cells will fill-in automatically if needed
5. Blue cells are merely for informational purposes
6. Click on the yellow button to save data to the "Statistics" tab if desired
  - \* This will create a running log of the calculated values for each test entered
7. Click on the "TST Summary Sheet" tab for a printer friendly version of the data and print if desired
8. Click on the green button on the "Calculator" tab to clear all the red input cells to enter new dataset
  - \* The same WET test with multiple endpoints (e.g., survival and reproduction) must be entered separately
  - \* Clearing the data on the "Calculator" tab will also clear the "TST Summary Sheet" - be sure to print before clearing
9. Enter your next set of test data as directed above and a log of your data will be formed on the "Statistics" tab
10. You must save the Excel workbook file when finished if you want your data to be available the next time you open the file

Results description:

A t-value is calculated for your dataset ("Calculated t-value") and compared against the "Table t-value" based on the degrees of freedom of your dataset. If the Calculated t-value is greater than the Table t-value, then the sample is not toxic at that concentration. If the calculated value is less than or equal to the table value, then the sample is toxic. In the event that there is no variance, the percent difference is used.

This tool was created for several of the EPA WET test methods for the West Coast marine species (USEPA 1995) and the 2002 EPA WET test methods (USEPA 2002a, 2002b, 2002c) for other species.

For questions, please contact:

Jaimie Gilliam  
Tetra Tech, Inc.  
410-356-8983



# TST Summary Sheet

<b>Lab Name</b>	USEPA Region 9 Laboratory	<b>Client Name</b>	Eugene Bromley, WTR-5
<b>Test ID</b>	Platform B	<b>Test Species</b>	<i>H. rufescens (red abalone)</i>
<b>Test Date</b>	4/15/2015	<b>Test Type</b>	Chronic
<b>Test Duration</b>	48 hr	<b>Endpoint</b>	Larval Development
<b>Critical Conc.</b>	0.0632		

Statistic	Control	Critical Concentration
Percent Mean of Raw Data	0.99	1.00
Mean used in Calculation (transformed)	1.46	1.51
Variance used in Calculation (transformed)	0.006	0.000
Standard Deviation of Transformed Data	0.077	0.021
CV of Transformed Data	0.052	0.014
n	10	10

### Mean % Effect at Critical Conc.

-1.32

Calculated t-value	Degrees of Freedom	Table t-value	Percent Difference
21.3456	11	1.7959	

### Results

Pass                      Sample is Non-toxic

### Raw Data

Control Data		Critical Concentration Data	
No. of Organisms Exposed or Counted	Response (Final Count, Weight, Length, etc.)	No. of Organisms Exposed or Counted	Response (Final Count, Weight, Length, etc.)
100	97	100	100
100	94	100	100
100	100	100	99
100	97	100	99
100	97	100	100
100	100	100	100
100	100	100	100
100	100	100	100
100	100	100	100
100	100	100	100
100	100	100	100



Statistics

<b>Lab Name</b>	USEPA Region 9 Laboratory	<b>Client Name</b>	Eugene Bromley, WTR-5	<b>Test ID</b>	B	<b>Test Date</b>	4/15/2015	<b>Test Duration</b>	48 hr	<b>Critical Conc.</b>	0.0632	<b>Test Species</b>	H. rufescens (red abalone)	<b>Chronic or Acute Endpoint</b>	Chronic Larval Development	<b>Mean % Effect at Critical Conc.</b>	-1.32	<b>Calculated t-value</b>	21.3456018	<b>Table t-value</b>	1.79588482	<b>Pass or Fail</b>	Pass	<b>Toxic or Non-toxic</b>	Non-toxic
-----------------	---------------------------	--------------------	-----------------------	----------------	---	------------------	-----------	----------------------	-------	-----------------------	--------	---------------------	-------------------------------	----------------------------------	----------------------------	--	-------	---------------------------	------------	----------------------	------------	---------------------	------	---------------------------	-----------



# TST Calculator v1.8

## TST Summary

The Test of Significant Toxicity (TST) is a new statistical approach that assesses the whole effluent toxicity (WET) measurement of wastewater effects on specific test organisms' ability to survive, grow, and reproduce developed by the U.S. EPA. TST uses hypothesis testing techniques which examines whether a sample, at the critical concentration (e.g., in-stream waste concentration or IWC), and the control within a WET test differ by an unacceptable amount (the amount that would have a measured adverse effect on the ability of aquatic organisms to thrive and survive). A sample can be either an effluent, stormwater, or receiving water. Once the WET test has been conducted, the TST approach can be used to analyze valid WET test results to assess whether the sample is toxic. The TST approach is designed to be used for a two concentration data analysis of the sample's critical concentration (e.g., IWC or a receiving water concentration) as compared to a control concentration.

## Instructions

Before using this program, Macros must be enabled and the security certificate must be accepted.

For Excel 2007

1. Click the "Office Button" (big Office logo in the upper left), and select "Excel Options" (lower right)
2. Click "Trust Center" on the left menu
3. Click "Trust Center Settings" (lower right)
4. Click "Macro Settings"
5. Click "Enable all macros" (last option)
6. Click "ok" buttons to clear all open windows
7. Exit program and re-open the file to use

For Excel pre-2007

1. Go to "Tools", "Macro", and Click "Security"
2. Set Macro security to medium
3. Exit program and re-open file
4. Click "Enable Macros"

To use the program:

1. Enter test information in the first block of red shaded cells (Test ID, Test Date, Test Duration, and Critical Concentration)
  - \* "Critical Conc." means a facility specific IWC or 100% receiving water, etc. (i.e., one concentration of WET test)
2. Fill in your raw test data (Number of Organisms Exposed or Counted and Response) for as many replicates as conducted in WET test
  - \* "Critical Conc. Data" means data from the concentration entered as the "Critical Conc." in cell B7
  - \* Percent data (survival, germination, etc.) should be input as whole numbers (not as decimals)
  - \* Fish growth must be input as biomass
3. Choose the appropriate test information from the red "Click to choose from menu" cells
4. All gray cells will fill-in automatically if needed
6. Click on the yellow button to save data to the "Statistics" tab if desired
  - \* This will create a running log of the calculated values for each test entered
7. Click on the "TST Summary Sheet" tab for a printer friendly version of the data and print if desired
8. Click on the green button on the "Calculator" tab to clear all the red input cells to enter new dataset
  - \* The same WET test with multiple endpoints (e.g., survival and reproduction) must be entered separately
  - \* Clearing the data on the "Calculator" tab will also clear the "TST Summary Sheet" - be sure to print before clearing
9. Enter your next set of test data as directed above and a log of your data will be formed on the "Statistics" tab
10. You must save the Excel workbook file when finished if you want your data to be available the next time you open the file

Results description:

A t-value is calculated for your dataset ("Calculated t-value") and compared against the "Table t-value" based on the degrees of freedom of your dataset. If the Calculated t-value is greater than the Table t-value, then the sample is not toxic at that concentration. If the calculated value is less than or equal to the table value, then the sample is toxic. In the event that there is no variance the percent difference is used.

This tool was created for several of the EPA WET test methods for the West Coast marine species (USEPA 1995) and the 2002 EPA WET test methods (USEPA 2002a, 2002b, 2002c) for other species.

For questions, please contact:

Jaime Gilliam  
Tetra Tech, Inc.  
410-356-8993



# TST Summary Sheet

<b>Lab Name</b>	USEPA Region 9 Laboratory	<b>Client Name</b>	Eugene Bromley, WTR-5
<b>Test ID</b>	Platform A	<b>Test Species</b>	<i>H. rufescens (red abalone)</i>
<b>Test Date</b>	4/15/2015	<b>Test Type</b>	Chronic
<b>Test Duration</b>	48 hr	<b>Endpoint</b>	Larval Development
<b>Critical Conc.</b>	0.0550		

Statistic	Control	Critical Concentration
Percent Mean of Raw Data	0.99	1.00
Mean used in Calculation (transformed)	1.47	1.51
Variance used in Calculation (transformed)	0.002	0.002
Standard Deviation of Transformed Data	0.048	0.039
CV of Transformed Data	0.033	0.026
n	10	10

### Mean % Effect at Critical Conc.

-0.91

Calculated t-value	Degrees of Freedom	Table t-value	Percent Difference
24.1193	17	1.7396	

### Results

Pass                      Sample is Non-toxic

### Raw Data

Control Data		Critical Concentration Data	
No. of Organisms Exposed or Counted	Response (Final Count, Weight, Length, etc.)	No. of Organisms Exposed or Counted	Response (Final Count, Weight, Length, etc.)
100	100	100	97
100	100	100	100
100	99	100	100
100	99	100	100
100	98	100	100
100	98	100	100
100	99	100	100
100	100	100	100
100	99	100	100
100	96	100	100



Statistics

<b>Lab Name</b>	<b>Client Name</b>	<b>Test ID</b>	<b>Test Date</b>	<b>Test Duration</b>	<b>Critical Conc.</b>	<b>Test Species</b>	<b>Chronic or Acute Endpoint</b>	<b>Mean % Effect at Critical Conc.</b>	<b>Calculated t-value</b>	<b>Table t-value</b>	<b>Pass or Fail</b>	<b>Toxic or Non-toxic</b>
USEPA Region 9 Laboratory	Eugene Bromley, WTR-5	Platform A	4/15/2015	48 hr	0.0550	H. rufescens (red abalone)	Larval Development	-0.91	24.1192899	1.73960673	Pass	Non-toxic

